

SEQ ID NO:1:

GGCATATTAGCTTGGGTTACTGTGAATTTTCTGACAGGTCAGCTGCATGGCCACAGA
CAGGAGACTGTGGGGACCTTGGACCTAGGGGGAGCCTCCACCCAAATCACGTTCT
GCCCCAGTTTGAGAAAACCTCTGGAACAACCTCCTAGGGGCTACCTCACTTCCTTTGA
GATGTTTAAACAGCACTTATAAGCTCTATACATAGTTACCTGGGATTTGGATTGAA
AGCTGCAAGACTAGCAACCCTGGGAGCCCTGGAGACAGAAGGGACTGATGGGCACA
CTTTCGGGAGTGCCTGT

SEQ ID NO:2:

GCGGGCTGCCGCGCAAGGGTGGCGCGCGCGCGTTCCTTGTTCTGGTCAACAAAG
AAATGTGGAGTGTCTTGGCTGAATCCTCATACAGACAAGATCATTATGGTGCTGTTA
GGTTGAAAAAGTGATATAATAAAGGAACCAAGGAGAAAATTCAGAAGGAAAGAAA
AAATTGCCTCTGCAGGTGTGCGAGCAGGATTGCTTCTGCAACAAAAGCCTCCACCCA
GCCACATCTTGGGAAAAGAATGGCCACTTCTTGGGGCACAGTCTTTTTCATGCTGGT
GGTATCCTGTGTTTGCAGCGCTGTCTCCACAGGAACCAGCAGACTTGGTTTGAGGG
TATCTTCCTGTCTTCCATGTGCCCCATCAATGTCAGCGCCAGCACCTTGTATGGAATT
ATGTTTGATGCAGGGAGCACTGGAACCTCGAATTCATGTTTACACCTTTGTGCAGAAA
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GCCTCCACCCAAATCACGTTCTTGGCCCCAGTTTGAGAAAACCTCTGGAACAACTCCT
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GAGGTGGGCTTTGAGCCCTGCTATGCCGAAGTGCTGAGGGTGGTACGAGGAAAAC
TCACCAGCCAGAGGAGGTCCAGAGAGGTTCTTCTATGCTTTCTCTTACTATTATGA
CCGAGCTGTTGACACAGACATGATTGATTATGAAAAGGGGGGTATTTTAAAAGTTGA
AGATTTTGAAAGAAAAGCCAGGGAAGTGTGTGATAACTTGGAAAACCTCACCTCAG
GCAGTCCTTTCCTGTGCATGGATCTCAGCTACATCACAGCCCTGTAAAGGATGGCT
TTGGCTTTGCAGACAGCACAGTCTTACAGCTCACAAAGAAAGTGAACAACATAGAG
ACGGGCTGGGCCTTGGGGGCCACCTTTCACCTGTTGCAGTCTCTGGGCATCTCCCAT
TGAGGCCACGTACTTCCTTGGAGACCTGCATTTGCCAACACCTTTTTAAGGGGAGGA
GAGAGCACTTAGTTTCTGAACTAGTCTGGGGACATCCTGGACTTGAGCCTAGAGATT
WRGTTAATTAASCGGCCGAGCTTATCCTTWATRAGGTAATTTACTTGCMTGGCCGCG
TTTACACGTCGTGATGGNAAACCTGCGTCCCAACTAACGCTTGASAMATCCCCTTCG
CAGCTGCGATACCAAAGCCGACGACGCCTTCCACAGTGCCA

Figure 1

SEQ ID NO:3.

MATSWGTVFFMLVVSCVCSAVSHRNQQTWFEGIFLSSMCPINVSASTLYGIMFDAGSTG
TRIHVYTFVQKMPGQLPILEGEVFDVSKPGLSAFVDQPKQGAETVQGLLEVAKDSIPRSH
WKKTPVVLKATAGLRLLPEHKAKALLFEVKEIFRKSPFLVPKGSVSIMDGSDEGILAWV
TVNFLTGQLHHRQETVGTLDLGGASTQITFLPQFEKTLEQTPRGYLTSEMFNSTYKLY
THSYLGFGGLKAARLATLGALETEGTDGHTFRSACLPRWLEAEWIFGGVKYQYGGNQEG
EVGFEP CYAEVLRVVVRGKLHQPEEVQRGSFYAFSYYYDRAVD TDMIDYEKGGILKVED
FERKAREVCDNLENFTSGSPFLCMDLSYITALLKDGFGFADSTVLQLTKKVNNIETGWA
LGATFHLLQSLGISH

Figure 2

Figure 3

1	M A T S W G T V F E F L V V S C V C S A V S H R N Q C T W F E C I F L A S M C P	246	prot
1	M A T S W G A V F - I M L A I A C V Q S T V F Y I R E Q C T W F E C I F L A S M C P	mur	ntpase
41	I N V S A L S T T Y G I M F D A G S T G T R I H V Y T F V Q A M R Q Q L P F L E C	246	prot
40	I N V S A Q T F Y G I M F D A G S T G T R I H V Y T F V Q A M R Q Q L P F L E C	mur	ntpase
81	E V F D S V K P G L S A F V D Q P R Q G A E T V Q C L L E V A K D S I P R S A W	246	prot
80	E I F D S V K P G L S A F V D Q P R Q G A E T V Q C L L E V A K D S I P R S A W	mur	ntpase
121	K I K T P V V L K A T A G L R L L P E H I K A K I A L L E I E V K E I P R K S P F L V P	246	prot
120	E R I T P V V L K A T A G L R L L P E Q K A C A L L E V E E I P K N S P F L V P	mur	ntpase
161	K G S V S I M D G S D E G I L A W V T V N F L T G Q L H G H R Q C E T V G T L D L	246	prot
50	D G S V S I M D G S Y E G I L A W V T V N F L T G Q L H G H R Q C E T V G T L D L	mur	ntpase
201	G G A S T Q I T F L P Q F E K T L E Q T P R G Y L T S F E M F N S T I Y K L Y T H	246	prot
200	G G A S T Q I T F L P Q F E K T L E Q T P R G Y L T S F E M F N S T I F K L Y T H	mur	ntpase
241	S Y L G F G L K A A R L A T L G A L E T E G T D G H T F R S A C L P R W L E A E	246	prot
240	S Y L G F G L K A A R L A T L G A L E A K G T D G H T F R S A C L P R W L E A E	mur	ntpase
281	W I F G G V K Y Q Y G G N Q E G E V G F E P C Y A E V L R V V R G K L H Q P E E	246	prot
280	W I F G G V K Y Q Y G G N Q E G E M I G F E P C Y A E V L R V V O G K L H Q P E E	mur	ntpase
321	V Q R G S F Y A F S Y Y Y D R A V I D T D M I D Y E K G G I L K V E D F E R K A R	246	prot
320	V R G S A F Y A F S Y Y Y D R A A D T H L I D Y E K G G V L K V E D F E R K A R	mur	ntpase
361	E V C D N L E N F T S G S P F L C M D L S Y I T A L L K D G F G F A D S T V L I Q	246	prot
360	E V C D N L G S F S S G S P F L C M D L T Y I T A L L K D G L G F A E R H P L T	mur	ntpase
401	L T K K V N N I E T G W - A L G A T F - - - - - H L L O S L G I S H	246	prot
400	- A H K E S E Q H R D W L G L G G H L S P A P V S G H H Q L R P S S T S E A C I	mur	ntpase
428		246	prot
439	S E P V F S O E G V D S E T F S D L S G K A W P E T R	mur	ntpase

Figure 4

Apyrase Conserved Regions in CD39-L4

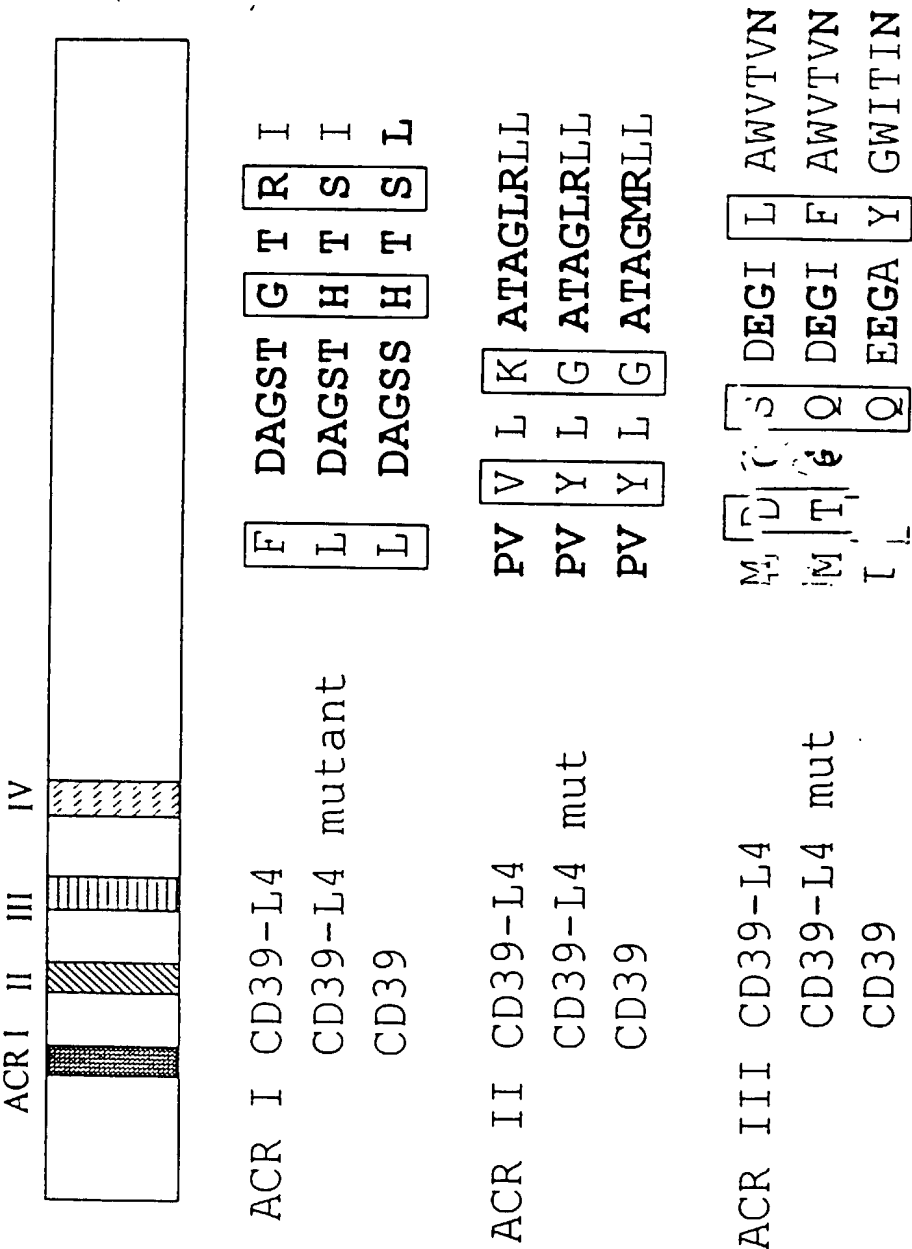


FIG. 5

10032047 . 020602

Nucleotide sequence of the CD39-L4 mutant ACRIII (SEQ ID NO:6). The nucleotide changes have been highlighted. The G to A and A to C changes at positions 502 and 503 produce a Thr, the T to C, C to A and C to A changes at positions 508-510 result in a Gln and the A to C changed at position 525 result in a Phe.

```
ATGGCCACTTCTTGGGGCACAGTCTTTTTTCATGCTGGTGGTATCCTGTGTTTGCAGCGCTGTCT
CCCACAGGAACCAGCAGACTTGGTTTGAGGGTATCTTCCTGTCTTCCATGTGCCCCATCAATGT
CAGCGCCAGCACCTTGTATGGAATTATGTTTGATGCAGGGAGCACTGGAATCGAATTCATGTT
TACACCTTTGTGCAGAAAATGCCAGGACAGCTTCCAATTCTAGAAGGGGAAGTTTTTGTATTCTG
TGAAGCCAGGACTTTCTGCTTTTGTAGATCAACCTAAGCAGGGTGCTGAGACCGTTCAAGGGCT
CTTAGAGGTGGCCAAAGACTCAATCCCCCGAAGTCACTGGAAAAAGACCCAGTGGTCTTAAAG
GCAACAGCAGGACTACGCTTACTGCCAGAACACAAAGCCAAGGCTCTGCTCTTTGAGGTAAAGG
AGATCTTCAGGAAGTCACCTTTCCTGGTACCAAAGGGCAGTGTTAGCATCATGACTGGACAAGA
CGAAGGCATATTCGCTTGGGTTACTGTGAATTTTCTGACAGGTCAGCTGCATGGCCACAGACAG
GAGACTGTGGGGACCTTGGACCTAGGGGGAGCCTCCACCCAAATCACGTTCTTGCCCCAGTTTG
AGAAAACCTCTGGAACAAACTCCTAGGGGCTACCTCACTTCTTTGAGATGTTTAACAGCACTTA
TAAGCTCTATACACATAGTTACCTGGGATTTGGATTGAAAGCTGCAAGACTAGCAACCTGGGA
GCCCTGGAGACAGAAGGGACTGATGGGCACACTTCCGGAGTGCCTGTTTACCGAGATGGTTGG
AAGCAGAGTGGATCTTTGGGGGTGTGAAATACCAGTATGGTGGCAACCAAGAAGGGGAGGTGGG
CTTTGAGCCCTGCTATGCCGAAGTGCTGAGGGTGTTACGAGGAAAACCTCACCAGCCAGAGGAG
GTCCAGAGAGGTTCCCTTCTATGCTTCTTCTTACTATTATGACCGAGCTGTTGACACAGACATGA
TTGATTATGAAAAGGGGGGTATTTTAAAAGTTGAAGATTTTGAAAGAAAAGCCAGGGGAAGTGTG
TGATAACTTGGAACCTTCACC TCAGGCAGTCCCTTCTGTGCATGGATCTCAGCTACATCAC
AGCCCTGTTA AAGGATGGCTTTGGCTTTGCAGACAGCACAGTCTTACAGCTCACAAAGAAAGT
GAAC AACATAG AGACGGGCTGGGCCTTGGGGGCCACCTTTCACCTGTTGCAGTCTCTGGGCA
TCTCCATTGA
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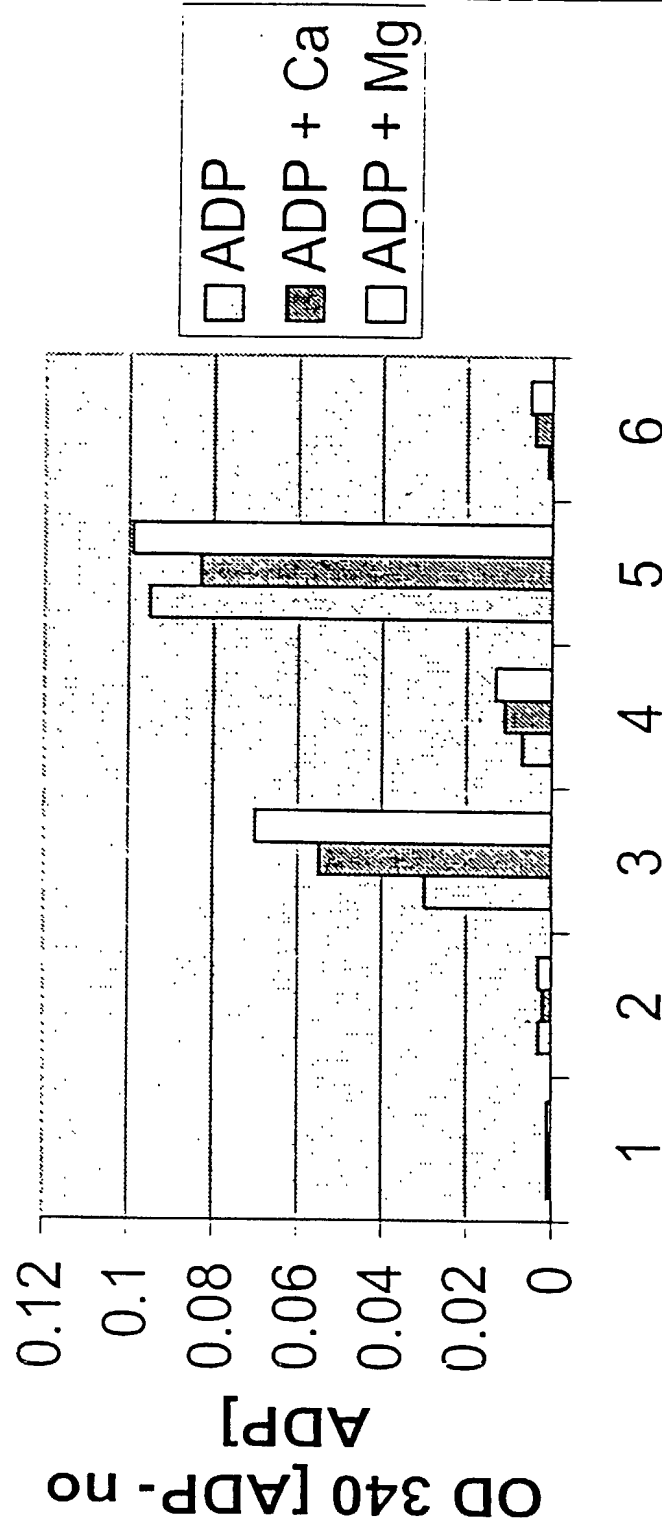
Amino acid sequence of CD39-L4 mutant ACR III (SEQ ID NO:7). The amino acid changes are D to T (a.a. 168), S to Q (a.a. 170) and L to F (a.a. 175). The changes are shown in bold.

```
MATSYGTVFFMLVVSCVCSAVSHRNQQTWFEGIFLSSMCPINVSASTLYGIMFDAGSTGT
RIHVYTFVQKMPGQLPILEGEVFDVSVKPGLSAFVDQPKQGAETVQGLLEVAKDSIPRSHW
KKTPVVLKATAGLRLLPEHKAKALLFEVKEIFRKSPFLVPKGSVSIMTGQDEGIFAWVTV
NFLTGQLHGHQRQETVGTLDLGGASTQITFLPQFEKTLEQTPRGYLTSFEMFNSTYKLYTH
SYLGFGFLKAARLATLGALETEGTDGHTFRSACLPRWLEAEWIFGGVKYQYGGNQEGEVGF
EPCYAEVLRVVRGKLHQPEEVQRGSFYAFSYYDRAVDTDMDIDYEKGGILKVEDFERKAR
EVCNLENFTSGSPFLCMDLSYITALLKDGFGFADSTVLQLTKKVNNIETGVALGATFHL
LQSLGISH
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FIG. 6

10092063, 030502

**FIG. 2 ADPase activity of CD39-L4
 ACR mutants**



1) ACR I; 2) ACR II; 3) ACR III; 4)
 Wild Type; 5) CD39; 6) Vector

CD39-L4

FIG. 7

Figure 8

SEQ ID No-3	M A T S W G	1	- - - - -	- V F F M L V V S C V C S A V S H R N Q Q T W F E G I F L S S M C P I N V S A S	46
SEQ ID No-5	M A T S W G T	- - - - -	- V F F M L V V S C V C S A V S H R N Q Q T W F E G I F L S S M C P I N V S A S		46
CD39Human.seq	M E D T K E S N V K T F C S K N I L A I L G F S	S I I A V I A	- - - - -	- L L A V G L T Q - - N K A L P E N	46
SEQ ID No-3	T L Y G I M F D A G S T G T R I H V Y T F V Q K M P G Q L P I L E G E V F D S V K	- P G L S A F V D Q P K Q G			100
SEQ ID No-5	T L Y G I M F D A G S T G T R I H V Y T F V Q K M P G Q L P I L E G E V F D S V K	- P G L S A F V D Q P K Q G			100
CD39Human.seq	V K Y G I V L D A G S S H T S L Y I Y K W P A E K E N D T G V V H Q V E E C R V K	G P G I S K F V Q K V N E I			101
SEQ ID No-3	A E T V Q G L L E V A K D S I P R S H W K K T P V V L K A T A G L R L L	- - P E H K A K A L L F E V K E I F			152
SEQ ID No-5	A E T V Q G L L E V A K D S I P R S H W K K T P V V L K A T A G L R L L	- - P E H K A K A L L F E V K E I F			152
CD39Human.seq	G I Y L T D C M E R A R E V I P R S Q H Q E T P V Y L G A T A G M R L L	R M E S E L A D R V L D V V E R S L			156
SEQ ID No-3	R K S P F L V P K G S V S I M D G S D E G I L A W V T V N F L T G Q L	- - - - -	- - - - -	- H G H R Q E T V	195
SEQ ID No-5	R K S P F L V P K G S V S I M D G S D E G I L A W V T V N F L T G Q L	- - - - -	- - - - -	- H G H R Q E T V	195
CD39Human.seq	S N Y P F D F Q - - G A R I I T G Q E E G A Y G W I T I N Y L L G K F S Q K T R W F S I V P Y E T N N Q E T F				209
SEQ ID No-3	G T L D L G G A S T Q I T F L P Q F E K T L E Q T P R G Y L T S F E M F N S T Y K L Y T H S Y L G F G L K A A				250
SEQ ID No-5	G T L D L G G A S T Q I T F L P Q F E K T L E Q T P R G Y L T S F E M F N S T Y K L Y T H S Y L G F G L K A A				250
CD39Human.seq	G A L D L G G A S T Q V T F V P Q - N Q T I E S P D N A - - L Q F R L Y G K D Y N V Y T H S F L C Y G K D Q A				261
SEQ ID No-3	- - - R L A T L G A L E T E G	- - - - -	- - - - -	- T D G H T F R S A C L P R W L E A E W I F G G V K	287
SEQ ID No-5	- - - R L A T L G A L E T E G	- - - - -	- - - - -	- T D G H T F R S A C L P R W L E A E W I F G G V K	287
CD39Human.seq	L W Q K L A K D I Q V A S N E I L R D P C F H P G Y K K V V N V S D L Y K T P C T K R - F E M T L P F Q Q F -				314
SEQ ID No-3	Y Q Y G G N Q E G E V G F E P C Y A E V L R V V R G K	- - - - -	- - - - -	- L H Q P E E V Q R G S F Y A F S	330
SEQ ID No-5	Y Q Y G G N Q E G E V G F E P C Y A E V L R V V R G K	- - - - -	- - - - -	- L H Q P E E V Q R G S F Y A F S	330
CD39Human.seq	- - - - - E I Q I G N Y Q Q C H Q S I L E L F N T S Y C P Y S Q C A F N G I F L P L Q G D F G A F S A F -				363
SEQ ID No-3	Y Y Y D R - - - A V D T D M I D Y E K G G I L K V E D F E R K A R E V C D N L E N F T S G S P - F L - - - C M				378
SEQ ID No-5	Y Y Y D R - - - A V D T D M I D Y E K G G I L K V E D F E R K A R E V C D N L E N F T S G S P - F L - - - C M				378
CD39Human.seq	- Y F V M K F L N L T S E K V S Q E K V T E M - M K K E C A Q P W E - - E I K T S Y A G V K E K Y L S E Y C F				414
SEQ ID No-3	D L S Y I T A L L K D G F G F A D S T	- - - - -	- - - - -	- V L Q L T K K V N N I E T G W	412
SEQ ID No-5	D L S Y I T A L L K D G F G F A D S T	- - - - -	- - - - -	- V L Q A A - - - - -	402
CD39Human.seq	S G T Y I L S L L Q G Y H F T A D S W E H I F I G K I Q G S D A G W T L G Y M L N L T	- - - - -	- - - - -	- - - - -	459
SEQ ID No-3	A L G A T F H L L Q S L G I S H	- - - - -	- - - - -	- - - - -	428
SEQ ID No-5	- - - - - - - - - - - V L R -	- - - - -	- - - - -	- - - - -	406
CD39Human.seq	- - - - - - - - - - - N M I P A E Q P L S T P L S H S T Y V F L M V L F S L V L F T V A I I G L L I F H P P				502

FIG 8

mur ntpase MATSWGA -MLIIACVGSSTVFRYEQQTWFEGVFLSSMCPINV GTFYGI MFDA 54
 SEQ ID No-3 MATSWG1 FFM LVVSCVCSAVSHRNQQTWFEGIFLSSMCPINV .STLYGI MFDA 55
 SEQ ID No-5 MATSWGTVFFMLV VSCVCSAVSHRNQQTWFEGIFLSSMCPINVSA STL YGI MFDA 55

mur ntpase GSTGTRI HVYTFVQKTAGQLPFL EGEI FDSVKPGLSAFVDQPKQGAETVQELLEV 109
 SEQ ID No-3 GSTGTRI HVYTFVQKMPGQLPILEGEVFD SVKPGLSAFVDQPKQGAETVQGLLEV 110
 SEQ ID No-5 GSTGTRI HVYTFVQKMPGQLPILEGEVFD SVKPGLSAFVDQPKQGAETVQGLLEV 110

mur ntpase AKDSIPRSHWERTPVV LKATAGLRLLPEQKALQLLEV EIFKN SPFLV P DGSVS 164
 SEQ ID No-3 AKDSIPRSHWKKTTPVV LKATAGLRLLPEHKKAKALLFEVKEI FRKSPFLV PKGSVS 165
 SEQ ID No-5 AKDSIPRSHWKKTTPVV LKATAGLRLLPEHKKAKALLFEVKEI FRKSPFLV PKGSVS 165

mur ntpase IMDGSYEGILAWVTVNFLTGGQLHGRGQETVGTLDLGGASTQITFLPQFEKTL EQT 219
 SEQ ID No-3 IMDGSDEGILAWVTVNFLTGGQLHGRGQETVGTLDLGGASTQITFLPQFEKTL EQT 220
 SEQ ID No-5 IMDGSDEGILAWVTVNFLTGGQLHGRGQETVGTLDLGGASTQITFLPQFEKTL EQT 220

mur ntpase PRGYLTSFEMFNSTFKLYTHSYLGFG LKAA RLATLGAL EAKGTDGHTFRSACLPR 274
 SEQ ID No-3 PRGYLTSFEMFNSTYKLYTHSYLGFG LKAA RLATLGAL ETEGTDGHTFRSACLPR 275
 SEQ ID No-5 PRGYLTSFEMFNSTYKLYTHSYLGFG LKAA RLATLGAL ETEGTDGHTFRSACLPR 275

mur ntpase WLEAEWIFGGVKYQYGGNQEGEMGFEP CYAEVLRV VQ GK LHQPEEV RGS AFYAFS 329
 SEQ ID No-3 WLEAEWIFGGVKYQYGGNQEGEVGFEP CYAEVLRV VRGK LHQPEEVQRGSFYAFS 330
 SEQ ID No-5 WLEAEWIFGGVKYQYGGNQEGEVGFEP CYAEVLRV VRGK LHQPEEVQRGSFYAFS 330

mur ntpase YYYDRAADTHLIDYEKGGV LKVEDFERKAREVCDNLG SFSSGSPFLC MDLTYITA 384
 SEQ ID No-3 YYYDRAVD TDMIDYEKGGILKVEDFERKAREVCDNLN ENFTSGSPFLC MDLTYITA 385
 SEQ ID No-5 YYYDRAVD TDMIDYEKGGILKVEDFERKAREVCDNLN ENFTSGSPFLC MDLTYITA 385

mur ntpase LLKDG LGFAERHPLT -AHK ESEQH R D W LG LGHLS PAVSGH HQLRPSSTSEACI 438
 SEQ ID No-3 LLKDGFGFADSTVLQLT KKVNNIETGW -ALGATF - - - - - H LQLSLGIS - - - 427
 SEQ ID No-5 LLKDGFGFADSTVLQ -A - - - - - - - - - - - - - - - AVL R - - - 405

mur ntpase SEPVSQEGVDSETFSDLSGKAWPETR 465
 SEQ ID No-3 - H 428
 SEQ ID No-5 - 406

FIG 9